PATENT COOPERATION TREATY
PCT
INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 11441PC2-MLE/AKB	**FOR FURTHER ACTION**	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416).
International Application No. **PCT/AU2003/001451**	International Filing Date *(day/month/year)* 3 November 2003	Priority Date *(day/month/year)* 7 November 2002

International Patent Classification (IPC) or national classification and IPC

Int. Cl. [7] C07K 7/06, 7/08, 14/05, 16/08, C07H 21/04, C12N 15/79, A61K 38/08, 38/10, 31/7088, A61P 31/20, G01N 33/53, 33/566, 33/68

Applicant

THE COUNCIL OF THE QUEENSLAND INSTITUTE OF MEDICAL RESEARCH et al

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 5 sheets, including this cover sheet.

   [X] This report is also accompanied by ANNEXES, i.e., sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of **12** sheet(s).

3. This report contains indications relating to the following items:

   - I [X] Basis of the report
   - II [ ] Priority
   - III [ ] Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV [ ] Lack of unity of invention
   - V [X] Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI [ ] Certain documents cited
   - VII [ ] Certain defects in the international application
   - VIII [X] Certain observations on the international application

Date of submission of the demand 11 March 2004	Date of completion of the report 28 February 2005
Name and mailing address of the IPEA/AU AUSTRALIAN PATENT OFFICE PO BOX 200, WODEN ACT 2606, AUSTRALIA E-mail address: pct@ipaustralia.gov.au Facsimile No. (02) 6285 3929	Authorized Officer **G. D. HEARDER** Telephone No. (02) 6283 2553

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No.
PCT/AU2003/001451

I. Basis of the report

1. With regard to the **elements** of the international application:*

☐ the international application as originally filed.

☒ the description, pages **1, 2, 4, 6-11, 15-50,** as originally filed,
pages , filed with the demand,
pages **3, 5, 12-14,** received on **2 August 2004** with the letter of **29 July 2004**

☒ the claims, pages , as originally filed,
pages , as amended (together with any statement) under Article 19,
pages , filed with the demand,
pages **51-57,** received on **2 August 2004** with the letter of **29 July 2004**

☒ the drawings, pages **1/11-11/11,** as originally filed,
pages , filed with the demand,
pages , received on with the letter of

☒ the sequence listing part of the description:
pages **1-33,** as originally filed
pages , filed with the demand
pages , received on with the letter of

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.
These elements were available or furnished to this Authority in the following language which is:

☐ the language of a translation furnished for the purposes of international search (under Rule 23.1(b)).

☐ the language of publication of the international application (under Rule 48.3(b)).

☐ the language of the translation furnished for the purposes of international preliminary examination (under Rules 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

☒ contained in the international application in written form.

☒ filed together with the international application in computer readable form.

☐ furnished subsequently to this Authority in written form.

☐ furnished subsequently to this Authority in computer readable form.

☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished

4. ☐ The amendments have resulted in the cancellation of:

☐ the description, pages

☐ the claims, Nos.

☐ the drawings, sheets/fig.

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).**

* *Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17).*

** *Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report*

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Claims	**1-6, 8, 10, 11, 13-17, 22, 25, 28, 29, 31-33, 37, 38, 40, 41**	**YES**
	Claims	**7, 9, 12, 18-21, 23, 24, 26, 27, 30, 34-36, 39, 42-54**	**NO**
Inventive step (IS)	Claims	**1-6, 8, 10, 11, 13-17, 22, 25, 28, 29, 31-33, 37, 38, 40, 41**	**YES**
	Claims	**7, 9, 12, 18-21, 23, 24, 26, 27, 30, 34-36, 39, 42-54**	**NO**
Industrial applicability (IA)	Claims	**1-54**	**YES**
	Claims		**NO**

2. Citations and explanations (Rule 70.7)

The following documents identified in the International Search Report have been considered for the purposes of this report:

D1 WO 1999/002550
D2 EP 1 229 043
D3 JP 2002-255997
D4 LEEN A et al.: .. Journal of Virology, September 2001, p 8649-8659
D5 THORLEY-LAWSON D A et al.: .. Proc. Nat. Acad. Sci. USA (1987), 84(15), 5384-8
D6 O'SULLIVAN D ET AL: .. J. of Immunology, Vol. 145, No. 6, September 15 1990, pp1799-1808
D7 MEIJ P et al.: .. International Journal of Cancer, 26 February 2002, 99(1), 93-99
D8 KHANNA R et al.: .. European Journal of Immunology (1998), 28(2), 451-458

Novelty

Claims 7, 9, 12, 18-21, 23, 24, 26, 27, 30, 34-36, 39, 42-54

D2 discloses a sequence SEQ ID NO 3 that renders SEQ ID NO 26 (ie claim 7, 9 (SEQ ID NO 23) and associated claims) not novel.

D3 discloses a sequence SEQ ID NO 19 that renders claim 9 (SEQ ID NOs 1, 22) and associated claims not novel. D3 also discloses a method that falls within the scope of claim 51 (and associated claims), see paragraphs [0014]-[0055].

D4 also discloses a method that falls within the scope of claim 51 (and associated claims).

D5 discloses a sequence LMP (158-167) that renders claim 9 (SEQ ID NO 1) and associated claims not novel.

D7 also discloses a method that falls within the scope of claim 51 (and associated claims).

Inventive Step

Claims 7, 9, 12, 18-21, 23, 24, 26, 27, 30, 34-36, 39, 42-54: as above.

Supplemental Box
(To be used when the space in any of the preceding boxes is not sufficient)

Continuation of Box V. 2. Citations and explanations

Non-patent Literature P Category Documents

The following documents may be relevant if there is a priority issue with the application:

D9 DURAISWAMY J et al: .. Journal of Virology, July 2003, pp 7401-7410

D10 DURAISWAMY J et al: .. Blood, 15 April 2003, 101(8), pp 3150-3156

VIII. Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

(a) Claim 13 is not clear because it is appended to itself.

(b) Claim 18 is not clear because the reference to "any one of Claims 1-1" does not make sense.

(iv) GGALLVLYSFAL (SEQ ID NO:35); and

(v) DWTGGALLVLYSFALML (SEQ ID NO:36).

In a fifth aspect, the invention provides an isolated EBV CTL peptide epitope comprising the amino acid sequence DSNSNE (SEQ ID NO:8), specifically excluding the amino acid sequence ESDSNSNEG (SEQ ID NO:2).

In a preferred embodiment of the fifth aspect, the epitope comprises an amino acid sequence selected from the group consisting of:

(i) DSNSNEGRH (SEQ ID NO:37);

(ii) SGHESDSNSNEG (SEQ ID NO:38); and

(iii) TDDSGHESDSNSNEGRH (SEQ ID NO:39).

The invention also provides a variant EBV CTL epitope.

In particular embodiments, said variant has an amino acid sequence set forth in Table 4 (SEQ ID NOS:40-44 and 47-50).

In a sixth aspect, the invention provides an isolated protein comprising at least one EBV CTL epitope according to any of the aforementioned aspects.

Preferably, the isolated protein is a polyepitope protein comprising an amino acid sequence selected from the group consisting of ALLVLYSFA (SEQ ID NO:30) and IALYQQNW (SEQ ID NO:21).

In a preferred embodiment, the isolated polyepitope protein comprises each of the EBV CTL epitopes set forth in Table 5.

In a particularly preferred embodiment, the isolated polyepitope protein comprises the amino acid sequence set forth in SEQ ID NO:81.

In a seventh aspect the invention an isolated nucleic acid encoding the EBV CTL epitope or polyepitope of any of the aforementioned aspects.

In a preferred embodiment, the isolated nucleic acid encodes the polyepitope amino acid sequence set forth in SEQ ID NO:81.

More preferably the isolated nucleic acid of this embodiment has the nucleotide sequence set forth in SEQ ID NO:82.

This aspect also provides an isolated nucleic acid encoding an EBV CTL epitope variant of the aforementioned aspects.

By "*EBV CTL epitope*" is meant a sequence of amino acids that is encoded by an EBV genome and is capable of eliciting an immune response by at least one T cell clonotype when the amino acid sequence is presented to the at least one T cell clonotype in the context of MHC class I *in vivo* or *in vitro*. This definition does not exclude T cell epitopes that, in addition, are T helper or B cell epitopes, for example.

The consensus amino acid sequences set forth in SEQ ID NOS:4-8 are minimal, common regions present in groups of EBV CTL peptides of the invention (SEQ ID NOS: 2 and 9-25, and 27-39) listed in Table 2.

Suitably, said EBV CTL peptide epitope consists of at least nine (9) and no more than twenty (20) contiguous amino acids.

In certain embodiments, said EBV CTL peptide epitope consists of nine (9), twelve (12) or seventeen (17) contiguous amino acids.

In a preferred embodiment, said EBV CTL peptide epitope has an amino acid sequence selected from the group consisting of: SEQ ID NO:2; SEQ ID NO:9; SEQ ID NO:10: SEQ ID NO:11; SEQ ID NO:12; SEQ ID NO:16; SEQ ID NO:17; SEQ ID NO:21; SEQ ID NO:22; SEQ ID NO:23; SEQ ID NO:24; SEQ ID NO:28; SEQ ID NO:29; SEQ ID NO:30; SEQ ID NO:31; and SEQ ID NO:37

The invention also contemplates isolated proteins, such as polypeptides or "polyepitope" proteins, comprising one or more, or preferably a plurality of, EBV CTL epitopes of the invention. For example, said epitopes may be present singly or as repeats, which also includes tandemly repeated epitopes. "Spacer" amino acids may also be included between one or more of the EBV CTL epitopes present in said isolated protein.

In one embodiment, an isolated protein may consist of one or more or, preferably, a plurality of, EBV CTL epitopes of the invention.

In another embodiment, an isolated protein may consist essentially of one or more or, preferably, a plurality of, EBV CTL epitopes of the invention.

By "*consist essentially of*" is meant in this context that the or each EBV CTL peptide epitope has no more than five or preferably no more three amino acids in addition to the EBV CTL epitope sequence.

In the particular context of a polyepitope protein, these additional amino acid residues may be referred to as "*spacer*" amino acids.

It will also be appreciated that polyepitope proteins of the invention may additionally comprise one or more other EBV CTL peptides, such as one or more of the prior art LMP epitopes described in Table 2 or Table 5 and/or the EBV CTL epitopes YLLEMLWRL (SEQ ID NO:2) andYLQQNWWTL (SEQ ID NO:1).

In a preferred embodiment, the isolated polyepitope protein comprises a plurality of MHC Class I-restricted LMP1 and/or LMP2 CTL epitopes.

Preferably, at least one of said CTL epitopes has an amino acid sequence selected from the group consisting of ALLVLYSFA (SEQ ID NO:30) and IALYQQNW (SEQ ID NO:21).

In a particularly preferred embodiment, the isolated polyepitope comprises thirteen EBV CTL epitopes having the respective amino acid sequences YLLEMLWRL (SEQ ID NO: 3); YLQQNWWTL (SEQ ID NO: 1); ALLVLYSFA (SEQ ID NO:30); IAYLQQNW (SEQ ID NO:21); SSCSSCPLSKI (SEQ ID NO: 51); PYLFWLAAI (SEQ ID NO:52); TYGPVFMCL (SEQ ID NO:53); RRRWRRLTV (SEQ ID NO:54); LLSAWILTA (SEQ ID NO: 55); LTAGFLIFL (SEQ ID NO:56); VMSNTLLSAW (SEQ ID NO:57); IEDPPFNSL (SEQ ID NO:58); CLGGLLTMV (SEQ ID NO:59).

Even more preferably, the isolated polyepitope protein has the amino acid sequence set forth in SEQ ID NO:81 (FIG. 8).

The CTL epitopes in the polyepitope in Table 5 and FIG. 8 have been selected to encompass a broad range of MHC Class I specificities. For example, it is estimated that these optimally-selected HLA specificities would encompass about 90% of the Asian, African and Caucasian populations.

It is also noted that the isolated polyepitope protein in FIG 8 (SEQ ID NO:81) has been shown by the present inventors to induce CD8+ CTL responses in polyepitope-immunized mice, which responses protect mice from tumour challenge.

Also contemplated are EBV CTL epitope variants.

Generally, as used herein, "*variants*" are EBV CTL epitopes of the invention in which one or more amino acids have been deleted or replaced by different amino acids without substantial alteration to immunogenicity. It is well understood in the art that some amino acids may be changed to others with broadly similar properties without changing the immunogenicity of the peptide (conservative substitutions).

Substantial changes in function are made by selecting substitutions that are less conservative and relatively fewer of these may be tolerated. Generally, the substitutions which are likely to produce the greatest changes in a protein's properties are those in which (a) a hydrophilic residue (e.g., Ser or Thr) is substituted for, or by, a hydrophobic residue (e.g., Ala, Leu, Ile, Phe or Val); (b) a cysteine or proline is substituted for, or by, any other residue; (c) a residue having an electropositive side chain (e.g., Arg, His or Lys) is substituted for, or by, an electronegative residue (e.g., Glu or Asp) or (d) a residue having a bulky side chain (e.g., Phe or Trp) is substituted for, or by, one having a smaller side chain (e.g., Ala, Ser)or no side chain (e.g., Gly).

Specific examples of naturally-occurring variant EBV CTL epitopes are provided in Table 4 and SEQ ID NOS:40-44 and 47-50, which variants are derived from particular ethnoregional EBV isolates and will be discussed in more detail hereinafter.

As will be evident from Table 4, examples of variant EBV CTL epitopes are provided which respectively differ from SEQ ID NO:1, SEQ ID NO:21 and SEQ ID NO:30 by a single amino acid. Variants of SEQ ID NO:3 differ by one, two or three amino acid residues.

The invention also contemplates "*derivatives*" of EBV CTL epitopes of the invention, such as created by chemical modification of amino acid residues, biotinylation, conjugation with fluorochromes, addition of epitope tags (for example c-*myc*, haemagglutinin and FLAG tags), and fusion partners that facilitate recombinant protein expression, detection and purification (such as glutathione-*S*-transferase, green fluorescent protein, hexahistidine and maltose-binding protein, although without limitation thereto).

CLAIMS

1. An isolated EBV CTL peptide epitope consisting of up to nine contiguous amino acid residues of an LMP1 protein, and comprising an amino acid sequence selected from the group consisting of:

(i) QRH (SEQ ID NO:4);

(ii) AGNDG (SEQ ID NO:5);

(iii) QNW (SEQ ID NO:6), specifically excluding the sequence YLQQNWWTL (SEQ ID NO:1);

(iv) VLYS (SEQ ID NO:7); and

(v) DSNSNE (SEQ ID NO:8),.

2. The isolated EBV CTL peptide epitope of Claim 1 consisting of an amino acid sequence selected from the group consisting of:

(i) QRHSDEHHH (SEQ ID NO:9);

(ii) GQRHSDEHH (SEQ ID NO:10);

(iii) YYHGQRHSD (SEQ ID NO:11); and

(iv) WMYYHGQRH (SEQ ID NO:12).

3. The isolated EBV CTL epitope of Claim 1 consisting of an amino acid sequence selected from the group consisting of:

(i) AGNDGGPPQ (SEQ ID NO:16); and

(ii) PSDSAGNDG (SEQ ID NO:17).

4. The isolated EBV CTL epitope of Claim 1 consisting of an amino acid sequence selected from the group consisting of:

(i) IALYLQQNW (SEQ ID NO:21);

(ii) ALYLQQNWW (SEQ ID NO:22);

(iii) QNWWTLLVD (SEQ ID NO:23); and

(iv) LYLQQNWWT (SEQ ID NO:24).

5. The EBV CTL peptide epitope of Claim 1 consisting of an amino acid sequence selected from the group consisting of:

(i) LLVLYSFAL (SEQ ID NO:29);

(ii) ALLVLYSFA (SEQ ID NO:30); and

(iii) VLYSFALML (SEQ ID NO:31).

6. The EBV CTL peptide epitope of Claim 1 consisting essentially of an amino acid sequence selected from the group consisting of:

(i) DSNSNEGRH (SEQ ID NO:37);and

(ii) ESDSNSNEG (SEQ ID NO:2).

7. An isolated EBV CTL peptide epitope consisting of an amino acid sequence selected from the group consisting of: ESDSNSNEG (SEQ ID NO:2); QRHSDEHHH (SEQ ID NO:9); GQRHSDEHH (SEQ ID NO:10); YYHGQRHSD (SEQ ID NO:11); WMYYHGQRH (SEQ ID NO:12); YYHGQRHSDEHH (SEQ ID NO:13); IWMYYHGQRHSD (SEQ ID NO:14); LIWMYYHGQRHSDEHHH (SEQ ID NO:15); AGNDGGPPQ (SEQ ID NO:16); PSDSAGNDG (SEQ ID NO:17); SDSAGNDGGPPQ (SEQ ID NO:18); DSAGNDGGPPQL (SEQ ID NO:19); PHSPSDSAGNDGGPPQL (SEQ ID NO:20); IALYLQQNW (SEQ ID NO:21); ALYLQQNWW (SEQ ID NO:22); QNWWTLLVD (SEQ ID NO:23); LYLQQNWWT (SEQ ID NO:24); IALYLQQNWWTL(SEQ ID NO:25); YLQQNWWTLLVD (SEQ ID NO:26); LIIALYLQQNWWTLLVD (SEQ ID NO:27); ALLVLYSFAL(SEQ ID NO:28); LLVLYSFAL (SEQ ID NO:29); ALLVLYSFA (SEQ ID NO:30); VLYSFALML (SEQ ID NO:31); ALLVLYSFALML (SEQ ID NO:32); GALLVLYSFALM (SEQ ID NO:33); DWTGGALLVLYS (SEQ ID NO:34); GGALLVLYSFAL (SEQ ID NO:35); DWTGGALLVLYSFALML (SEQ ID NO:36); DSNSNEGRH (SEQ ID NO:37); SGHESDSNSNEG (SEQ ID NO:38); and TDDSGHESDSNSNEGRH (SEQ ID NO:39).

8. An isolated EBV CTL peptide epitope consisting of an amino acid sequence selected from the group consisting of: SEQ ID NO:2; SEQ ID NO:9; SEQ ID NO:10: SEQ ID NO:11; SEQ ID NO:12; SEQ ID NO:16; SEQ ID NO:17; SEQ ID NO:21; SEQ ID NO:22; SEQ ID NO:23; SEQ ID NO:24; SEQ ID NO:28; SEQ ID NO:29; SEQ ID NO:30; SEQ ID NO:31; and SEQ ID NO:37

9. A variant of an isolated EBV CTL epitope that consists of an amino acid sequence sequence differing by one, two or three amino acids from an amino acid sequence selected from the group consisting of: SEQ ID NO:1; SEQ ID NO:2; SEQ ID NO:9; SEQ ID NO:10: SEQ ID NO:11; SEQ ID NO:12; SEQ ID NO:16;

SEQ ID NO:17; SEQ ID NO:21; SEQ ID NO:22; SEQ ID NO:23; SEQ ID NO:28; SEQ ID NO:29; SEQ ID NO:30; SEQ ID NO:31; and SEQ ID NO:37

10. The variant of Claim 9, comprising an amino acid sequence selected from the group consisting of:

(i) an amino acid sequence differing from SEQ ID NO:1 by one amino acid;

(ii) an amino acid sequence differing from SEQ ID NO:21 by one amino acid; and

(iii) an amino acid sequence differing from SEQ ID NO:30 by one amino acid.

11. A variant of an isolated EBV peptide epitope having an amino acid sequence according to any one of SEQ ID NOS:40-44 and 47-50.

12. An isolated protein comprising at least one EBV CTL epitope and/or EBV CTL epitope variant according to any one of Claims 1-11.

13. The isolated protein of Claim 13, comprising a plurality of EBV CTL epitopes and/or EBV CTL epitope variants.

14. The isolated protein of Claim 13 which is a polyepitope protein comprising an amino acid sequence selected from the group consisting of ALLVLYSFA (SEQ ID NO:30) and IALYQQNW (SEQ ID NO:21).

15. The isolated protein of Claim 14, wherein a plurality of the EBV CTL epitopes and/or EBV CTL epitope variants are contiguous.

16. The isolated polyepitope protein of Claim 15 comprising thirteen EBV CTL epitopes having the respective amino acid sequences YLLEMLWRL (SEQ ID NO: 3); YLQQNWWTL (SEQ ID NO: 1); ALLVLYSFA (SEQ ID NO:30); IAYLQQNW (SEQ ID NO:21); SSCSSCPLSKI (SEQ ID NO: 51); PYLFWLAAI (SEQ ID NO:52); TYGPVFMCL (SEQ ID NO:53); RRRWRRLTV (SEQ ID NO:54); LLSAWILTA (SEQ ID NO: 55); LTAGFLIFL (SEQ ID NO:56); VMSNTLLSAW (SEQ ID NO:57); IEDPPFNSL (SEQ ID NO:58); CLGGLLTMV (SEQ ID NO:59).

17. The isolated polyepitope protein of Claim 16 comprising the amino acid sequence set forth in SEQ ID NO:81.

18. An isolated nucleic acid encoding the isolated EBV CTL epitope of any one of Claims 1-1.

19. An isolated nucleic acid encoding the isolated protein of any one of Claims 12-17.

20. An isolated nucleic acid encoding the variant EBV peptide epitope of any one of Claims 9-11.

21. The isolated nucleic acid of Claim 20 comprising a nucleotide sequence as set forth in any one of SEQ ID NOS: 63-65, 67-69, 71-76 or 78-80.

22. The isolated nucleic acid of Claim 19 which comprises the nucleotide sequence set forth in SEQ ID NO:80.

23. An expression construct comprising the isolated nucleic acid of any one of Claims 18-22 operably linked to one or more regulatory nucleotide sequences in an expression vector.

24. The expression construct of Claim 23, which is adenovirus-based.

25. The expression construct of Claim 24, which encodes the amino acid sequence set forth in SEQ ID NO: 81.

26. A host cell or organism comprising the expression construct of Claim 23.

27. A pharmaceutical composition comprising at least one isolated EBV CTL peptide epitope according to any one of Claims 1-8 and/or at least one isolated EBV CTL peptide epitope variant according to any one of Claims 9-11, together with a pharmaceutically acceptable carrier, diluent or excipient.

28. The pharmaceutical composition of Claim 27 comprising an amino acid sequence selected from the group consisting of ALLVLYSFA (SEQ ID NO:30) and IALYQQNW (SEQ ID NO:21).

29. The pharmaceutical composition of Claim 27 comprising a polyepitope protein that comprises the amino acid sequence set forth in SEQ ID NO:81.

30. A pharmaceutical composition comprising the expression construct of Claim 23 together with a pharmaceutically acceptable carrier, diluent or excipient.

31. The pharmaceutical composition of Claim 30 comprising an expression construct that encodes an amino acid sequence selected from the group consisting of ALLVLYSFA (SEQ ID NO:30) and IALYQQNW (SEQ ID NO:21).

32. The pharmaceutical composition of Claim 29 comprising an expression construct that encodes a polyepitope protein having the amino acid sequence set forth in SEQ ID NO:81.

33. The pharmaceutical composition of Claim 32 comprising the nucleotide sequence set forth in SEQ ID NO:82.

34. The pharmaceutical composition of any one of Claims 27-33, which is an immunotherapeutic composition.

35. The pharmaceutical composition of Claim 34, which is a vaccine.

36. A method of therapeutically and/or prophylactically treating an EBV-associated disease, including the step of administering to an animal at least one isolated EBV CTL epitope according to any one of Claims 1-12.

37. The method of Claim 36 wherein the at least one epitope comprises an amino acid sequence selected from the group consisting of ALLVLYSFA (SEQ ID NO:30) and IALYQQNW (SEQ ID NO:21).

38. The method of Claim 35 wherein the at least one peptide epitope is a polyepitope protein that comprises the amino acid sequence set forth in SEQ ID NO:81.

39. A method of therapeutically and/or prophylactically treating an EBV-associated disease, including the step of administering to an animal the expression construct of Claim 23.

40. The method of Claim 39 wherein the expression construct encodes a polyepitope protein that comprises an amino acid sequence selected from the group consisting of ALLVLYSFA (SEQ ID NO:30) and IALYQQNW (SEQ ID NO:21).

41. The method of Claim 40 wherein expression construct comprises the nucleotide sequence set forth in SEQ ID NO:82.

42. The method of any one of Claims 36 to 41, wherein the EBV associated disease is selected from B and T cell non-Hodgkin's lymphomas, Hodgkin's disease, and lymphoepithelioma-like carcinomas.

43. The method of Claim 42, wherein the EBV associated disease is nasopharyngeal carcinoma (NPC).

44. The method of any one of Claims 36-43 wherein the animal is a mammal.

45. The method of Claim 44 wherein the mammal is a human.

46. The method of Claim 45 wherein one or more of the at least one EBV peptide epitopes is selected according to a HLA type of the human to be treated.

47. An antibody which binds an EBV CTL epitope according to any one of Claims 1-8 or the variant of any one of Claims 9-11 .

48. A method of determining whether an animal harbours, or has been exposed to, Epstein Barr Virus, said method including the step of contacting one or more T cells isolated from said individual with at least one EBV peptide epitope according to any one of Claims 1-8, whereby a response to the at least one EBV peptide epitopes by said one or more T cells indicates that the animal harbours, or has been exposed to, Epstein Barr Virus.

49. The method of Claim 48 wherein the animal is a mammal.

50. The method of Claim 49, wherein the animal is a human.

51. A method of identifying an EBV CTL epitope, said method including the steps of:

(i) producing a plurality of different peptides derived from an EBV LMP1 protein;

(ii) combining said one or more of said peptides with one or more T lymphocytes obtained from an EBV seropositive individual; and

(iii) measuring IFN-γ production by said one or more T lymphocytes in response to said one or more peptides, wherein production of IFN-γ above a reference amount is indicative of said one or more peptides having at least one EBV CTL epitope.

52. The method of Claim 51 further including the step (iv) of determining whether said one or more T lymphocytes produced at step (ii) lyses one or more EBV-infected target cells.

53. An isolated EBV CTL epitope when obtained by the method of Claim 52.

54. The isolated CTL epitope of Claim 53 which has an amino acid sequence selected from the group consisting of: SEQ ID NO:2; SEQ ID NO:9; SEQ ID NO:10: SEQ ID NO:11; SEQ ID NO:12; SEQ ID NO:16; SEQ ID NO:17; SEQ

ID NO:21; SEQ ID NO:22; SEQ ID NO:23; SEQ ID NO:24; SEQ ID NO:28; SEQ ID NO:29; SEQ ID NO:30; SEQ ID NO:31; and SEQ ID NO:37.